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<b>13. ABSTRACT (Maximum 200 words)</b>  The techniques are employed to measure the optical absorption and scattering factors of breast tissue: 1) A pulse time device in which light at lower power (50 $\mu$ W) laser diode is employed. The time delay in light propagation from the input to output fiber optic couplers is related to the scattering factor and the measured rate of decay of emerging photons is related to the absorption factor. 2) Phase modulation, which is more convenient and rapid, measures the phase shift of a high frequency oscillating light of approximately the same power and wavelength used in the pulse time method. Here, there is direct reading of the time delay which is characteristic of the scattering power when low frequency oscillation is employed. Contrast agents may be used to increase the tumor detection sensitivity. For example, the absorption of porphyrins at 620 nm and indocyanine green at 860 nm, afford possibilities to follow the time course of tumor uptake of optical contrast agents. Histopathological confirmation of diagnosis will be obtained and correlated with the optical and MRI results. Both the imaging and optical data obtained from this proposed project will facilitate research on using the optical properties of breast tissue to screen for early stage cancer.					
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*Lawrence J. Flynn, MD* 4/13/95  
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## Annual Report

**Contract:** US Army Med. R&D Command DAMD 17-93-C3071  
**Title:** "Breast Cancer: Detection, Diagnosis and Treatment" Project V:  
Early Detection Breast Cancer and Recurrence using Near Infrared  
Time Resolved Spectrophotometry"  
**Principal Investigator:** Lawrence Solin, MD  
**Co-Investigators:** Britton Chance, Ph.D.  
Susan Orel, MD

### INTRODUCTION

**Nature of the Problem.** The possibility of simultaneous optical and MRI examination of tumor bearing subjects prior to surgery and histopathology, affords a unique opportunity to validate the optical method on a large patient population of verified tumor bearing breasts. This study could afford a "gold standard" for other studies of breast tumor.

**Background.** The success of the TRS method over the first and second years of this breast tumor study, not only has yielded quantitative values of absorption factor ( $\mu_a$ ) and scattering factor ( $\mu_s'$ ) of near infrared optical radiation (670 to 850 nm), but also have afforded evidence of the specificity of the optical method for tumors located in normal breast tissue. Because the protocol involves the study of identified tumors in breasts scheduled for surgical procedures, and because gadolinium enhanced MRI is used as a gold standard of tumor localization and identification, a gold standard of tumor size and location was available in all examinations. The specificity factor in MRI examinations is the selective uptake of the contrast agent, gadolinium chelate with respect to the background normal tissue. In Year 1 the specificity for the detection of breast tumor, best obtained based upon the tumor's increased blood content, has afforded the first and important step towards a primary measure of specificity. This is achieved with an intrinsic or endogenous contrast based upon the increased vascularity of the rapidly growing portion of the tumor. The second specificity factor is based upon the data of the Folkman laboratory (1), that among the blood vessels serving the tumor with increased blood concentration with respect to the host tissue, at least a portion of these vessels will show enhanced leakage of small molecules into the extravascular space with respect to of the normal tissue. Thus, the amount of a blood delivered contrast agent present in the tumor exceeds not only that of the normal tissue by virtue of increased amount of blood vessels, but also, at least portions of the extravascular space will be occupied by the contrast agent in excess of that of the normal tissue. Thus the optical sensitivity for the presence of a tumor rests not only on the contrast agent enhancement of the extravascular spaces by injection of a contrast agent but also due to the excess blood in the tumor volume itself.

**Purpose.** The goal of Year 3 affords an in depth comparison of the specificity of two optical determinants with the gold standard of tumor location. This is provided by gadolinium enhanced MRI in patients in which surgical verification of the tumor location and histopathological identification of the tumor type are afforded by the patient selection routine. It is believed that only by this tightly controlled evaluation procedure can we justify the advantages of speed, efficacy, safety and of the simple and economic optical method for tumor location and identification.

### Methods of Approach

The specific aims provide for measurements with two distinct methods, pulse time (TRS) and phase modulation.

Patient Recruitment. The collaborative project directed by the PI and involving collaborations of Drs. Ernest Rosato and Michael Torsian with the organization skills of nurse Jean McDermott has provided opportunities for our team, Dr. Shoko Nioka, Ms. Shinyin Zhao and Dr. H. Liu to:

- 1) solicit subjects for this procedure over and above the scheduled MRI study,
- 2) utilize efficiently the optical tumor detection apparatus, and
- 3) provide data analysis and comparison with the MRI surgical and histopathological data.

## EXPERIMENTAL METHODS / RESULTS OBTAINED

Case Summary. The patient population for this study consists of three groups of patients: 1) patients undergoing definitive breast irradiation following breast-conservation surgery for early stage breast cancer; 2) patients with locally advanced or inflammatory breast cancer; and 3) patients with possible recurrence in the breast, following breast-conserving surgery plus definitive breast irradiation. The number of patients studied to date in Group 1 is 35, which is substantially greater than the target accrual of 30 by year 2. The number of patients in Group 2 is 2, and the number of patients in Group 3 is 2, both of which are less than the target accrual of 20 each by year 2.

The increased accrual for Group 1, with the decreased accrual for Groups 2 and 3, is due to the nature of the patient population seen in the Department of Radiation Oncology at the Hospital of the University over the 2 years of the study. Characteristics of the patients studied in Group 1 are listed in Table 1.

Table I. Group 1 Patients undergoing Definitive Breast Irradiation in Early Stage Breast Cancer

	Number	(%)
<b>T Stage<sup>1</sup></b>		
Tis	3	(8)
T1	24	(69)
T2	8	(23)
<b>Tumor Pathology</b>		
Intraductal	3	(8)
Invasive		
Ductal	27	(77)
Lobular	1	(3)
Ductal/Lobular	1	(3)
Medullary	1	(3)
Tubular	1	(3)
Mucinous	1	(3)
<b>Timing of Pre-radiation Study</b>		
Before excision	11	(31)
After excision (before re-excision)	10	(29)
After excision (no re-excision)	4	(11)
After rre-excision	10	(29)
<b>Number of Studies</b>		
Pre-radiation only	15	(43)
Pre-radiation and 1 post radiation	9	(26)
Pre-radiation and ≥ 2 post radiation	11	(31)

<sup>1</sup>American Joint Committee on Cancer: Manual for Staging of Cancer (4th addition). J.B. Lippincott Co., Phila., PA, 1992, pp. 149-154.

## DATA ANALYSIS

Figure 1 illustrates the oxygen saturation in breast cancer on the above mentioned population showing a distribution covering a range from the normal of 75% down to 50% and less. Taking normoxic to lie between 70 and 80% saturation, we conclude that 20% are normoxic, 50% are hypoxic and 25% are hyperoxic. On a biochemical basis we attribute the hypoxic - anoxic fraction to be rapidly growing, inadequately vascularized tumors and the hyperoxic to be necrotic tumors. Thus, while the radiation sensitivity of the normoxic - hyperoxic group would be high, only 20% would be "worth while" to irradiate and 50% would be less sensitive to irradiation. Thus, already the preliminary results show important results of clinical significance that indeed may determine outcome for radiation oncology.

Summary. Preliminary determinations of oxygen saturation in tumor bearing breasts give a wide range of distributions which show over 50% of the breasts to be in a hypoxic state and therefore resistant to radiation as compared with the smaller fractions in normoxic and hypoxic states.

## O<sub>2</sub> saturation distribution in breasts with cancer ( 23 patients)

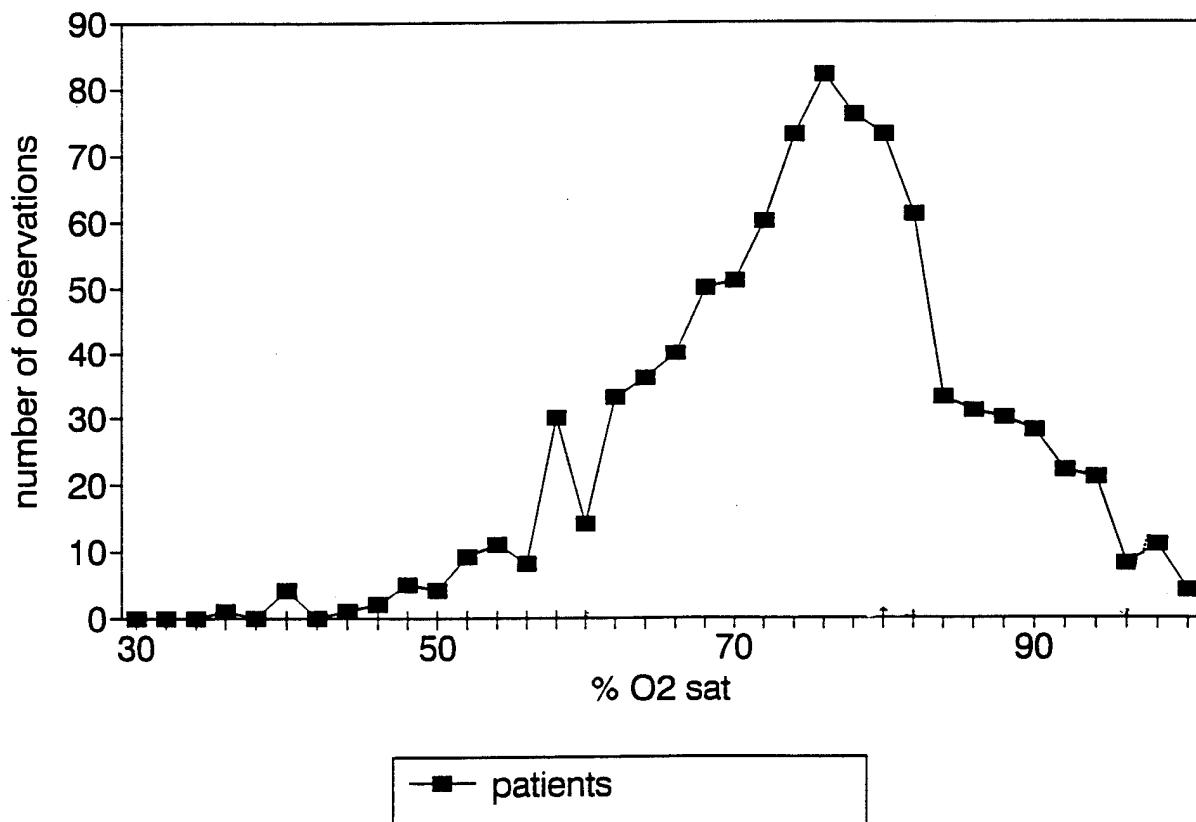


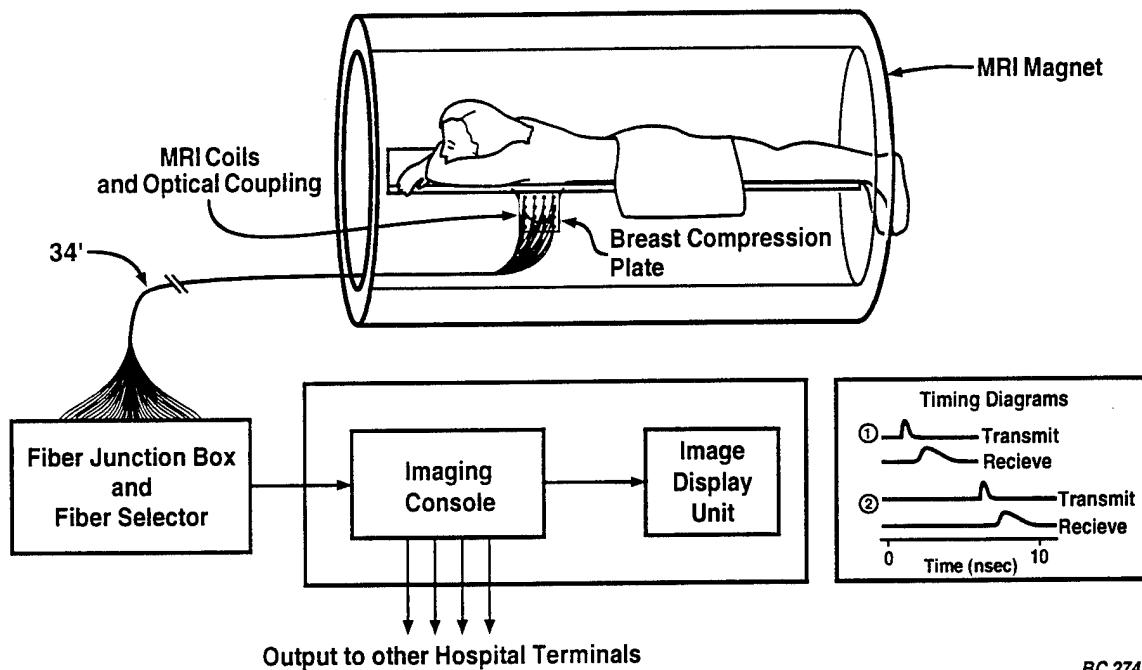
Figure 1. Oxygen saturation in tumor bearing breasts.

## Conclusions

The feasibility of time domain and phase modulation systems in the study of breast tumors is clearly demonstrated by Figure 1. Thus the feasibility study is completed and the two methods, time domain and phase modulation have shown efficacy in accumulating breast tumor data and are convenient and effective to use.

## Future Plans: Aims 2,3, and 4.

As is consistent with Aims 2, 3 and 4, we are proposing as a part of the Year 3 effort and as a main feature of any possible further funding to carry out the optical and MRI examinations simultaneously rather than consecutively since only a small fraction of the available population is willing remain to continue the study with the optical method. The diagram of Figure 2 illustrates our proposal to accomplish this by fiber optical coupling of the optical method directly into the magnet bore through 34 feet of 32 fibers, 16 on each side of the breast, and an additional 16 fibers on the contralateral breast as soon as the MRI has completed the "dual breast" coil assembly, leading to a potential for 64 optical fibers for coupling in and out of the MRI. Enthusiastic support of this project is evinced by the MRI team, particularly Dr. Schnall who has indicated that the soft compression plates used with the MRI are optically transparent and could readily be used to support at least 16 source detector fibers on each side of a single breast, or in fact, both breasts, because MRI plans to observe both breasts simultaneously in order to ensure that no tumors are located in the contralateral breast. Thus, sufficient fibers will be coupled directly into the MRI soft compression plates to afford tumor detection over the entire volume imaged by MRI, and thus ensure correlation of the results of the two methods. Furthermore, the two methods are non-interfering, and optical data can be acquired simultaneously with MRI data since no detectable magnetic field is induced by propagation of light through the optic fibers.



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Figure 2. Illustrating a method of procedure for fiber optic coupling of the time domain and phase modualtion optical method to the plastic breast compression plate used in the MRI studies. The diagram indicates the use of different fibers in the fiber optic coupling to obtain data from different places on the breast.

To ensure that none of the hardware of the optical system perturbs the magnetic field of the MRI, the optic fibers will be passed out through the electrostatic screen cage of the MRI over a distance of 34 feet to the optical console.

Given 16 source and 16 detector fibers, either time or frequency domain methods are feasible even though the pulse time, time domain device is illustrated in the diagram. Furthermore, most imaging algorithms can accept sparse data as would be provided by the availability of the 16 source detector combinations. However, our first use of the optical coupling will be with the TRS method. With TRS, the acquisition time is significantly diminished by virtue of the fact that a pair of eight new anode microchannel plate detector tubes will be used, together with a new three wavelength source laser diode system.

The time table for installation of in-magnet optical tumor detection: Other funds appear to be available for the installation of the fiber optic coupling and the services of this grant are required for the intensive clinical study that would ensure, approximately three breast examinations per week with a maximum of 8 or 10 on unusually busy weeks.

Staging of the Optical Study. The optical examination for increased blood concentration in the tumor region would afford the first step in the breast examination for tumor detection based upon the increased vascularity of the actively growing tumor. At the same time, the optical examination would permit study of light scattering increases to detect any increase of mitochondrial content of the tumor, with respect to background, and any increased scattering due to accumulation of calcium in the mitochondrial matrix leading to overt calcifications. The third step would be the intravenous injection of 0.5 mg/kg cardiogreen in order to enhance the specificity by this optical contrast agent. In all cases the two breasts would be compared in corresponding locations for their mitochondria and calcification light scattering and for the intensity and time course of their response to the IV contrast agent, cardiogreen.

Patient Consent. It is expected that those patients who have agreed to the MRI examination would not find it additionally stressful to have the optical examination to occur simultaneously since the total time of the examination would not be increased. In addition, since the intravenous injection of gadolinium chelate is an essential part of the MRI examination, it is expected that the additional injection of cardiogreen would not cause additional patient concern.

Timing of the Two Examinations. The optical examination can start any time that soft compression of the breast within the magnet is achieved, and indeed, regardless of whether the patient is in the proper position in magnet bore, or whether the shimming process has been completed or whether NMR data is actually being accumulated. The optical method is considered to be completely non-intrusive and thus the time available for the optical examination is the entire time from the initial prone position of the patient with compression plates in place until the end of the NMR examination when the compression is relieved.

Interaction of Gadolinium Chelate and Cardiogreen. The gadolinium chelate is designed to be completely unreactive within the body and is shown very clearly *in vitro* to be unreactive to cardiogreen. Thus, no interaction of the two *in vivo* is expected. The cardiogreen differs from the gadolinium chelate in that the cardiogreen is rapidly eliminated by the liver into the bile, whilst gadolinium chelate remains in the blood stream until it is finally eliminated in the kidney. Thus, the cardiogreen is expected to be eliminated into the bile duct prior to the completion of the MRI examination and not to be interfering in any possible way with the conduct of the former.

In summary, we regard simultaneous NMR and optical examination to be a significant factor in patient convenience and in the efficacy of the optical examination with no perceived additional risks for the patient study.

**IRB approval.** Application for IRB approval of the joint procedures is underway and is expected to be complete by the time budget year 3 is activated.

## **REFERENCES**

1. J. Folkman (1992) The Role of Angiogenesis in Tumor Growth. *Cancer Biol.* 3:65-71.